

IN THE CLAIMS

Before claim 1, change "Patent Claims" to --WHAT IS CLAIMED IS:--.

Delete claims 1-11.

Add the followings new claims 12-27:

12. (New) A wave field microscope comprising:

an illumination or excitation system having an object space and including in two or more spatial directions a first light beam source comprising at least one real or virtual illumination source for light beams capable of coherence and a second light beam source comprising at least one reflector or beam splitter for decoupling beam components or a further illumination source for light beams capable of coherence,

each of the first and second light beam sources being assigned at least one objective lens, and each being suited for generating light wave trains, the light wave trains of the first light beam source being aligned antiparallel or in variably adjustable angles to the light wave trains of the second light beam source such that the light wave trains emitted by the first light beam source interfere with those of the second light beam source to form a standing wave field having plane wave fronts; and

a detection system including at least one detection objective lens suitable for at least one of epifluorescent detection and raster scanning point detection, the detection objective lens being arranged with an optical axis normal to the plane wave fronts, and the detection objective lens being one of the at least one objective lens or another objective lens, the detection system also including a flat detector arranged upstream from the detection objective lens suitable for epifluorescent detection or for raster scanning point detection.

13. (New) The wave field microscope as recited in claim 12 wherein the flat detector is a camera capable of epiflorescent detection.

14. (New) The wave field microscope as recited in claim 12 wherein flat detector includes at least one of a stationary, confocal detection annular plate and aperture plate with at least one stationary detection slit being arranged upstream from it and further includes a point detector, in particular a photomultiplier, a photodiode, or a diode array being arranged downstream from the flat detector for raster scanning point detection.

15. (New) The wave field microscope as recited in claim 12 wherein in at least one spatial direction of the at least two spatial direction, an objective lens of a low numerical aperture or a reflector is assigned to an objective lens of a high numerical aperture, and in one or both other spatial directions, either two objective lenses of a low numerical aperture or an objective lens of a low numerical aperture and a reflector are assigned to one another.

16. (New) A wave field microscope comprising:

an illumination or excitation system having an object space and including in at least one of the three spatial directions a first illumination source including at least one real or virtual illumination source for light beams capable of coherence and at least one beam splitter for decoupling at least one beam component, and a common lens assigned to both the first illumination source and the at least one beam splitter into which light wave trains of the first illumination source and of the at least one beam splitter can be launched so as to produce on a rear focal plane facing away from the object space two spaced apart focal points, and that the light wave trains run relatively to each other in a variably adjustable angle in the space between the two focal planes, and interfere to form a one-dimensional, standing wave field; and

a detection system including at least one detection objective lens for at least one of epifluorescent detection and raster-scanning point detection, the at least one detection objective lens being one of the common lens and another lens, and further including a flat detector arranged upstream from the detection objective lens suited for epifluorescent detection or raster point detection.

17. (New) The wave field microscope as recited in claim 16 wherein the flat detector is a camera.

18. (New) The wave field microscope as recited in claim 16 wherein the flat detector is a raster-scanning point detector having at least one of a one stationary, confocal detection annular plate and aperture plate and at least one stationary detection slit arranged upstream, and further including a point detector including at least one of a photomultiplier, a photodiode, and a diode array arranged downstream.

19. (New) The wave field microscope as recited in claim 16 wherein the illumination or at least one further real or virtual illumination source for light beams or at least one further beam splitter for decoupling at least one beam further objective lens through which the light wave trains are focused into and are aligned in such away that they interfere with the light wave trains from the other or two other spatial direction so that the one or two-wave field form a two- or three-dimensional wave field.

The wave field microscope as recited in claim 12 in that the object space object mount fixture, in or on which an object is rotatably supported with g structures.

21. (New) The wave field microscope as recited in claim 20 further comprising at least one calibration target in the wave field, the object capable of being a rotated 360 degrees for at least one axis.

22. (New) The wave field microscope as recited in claim 12 wherein the illumination sources producing the multi-dimensional wave field, and/or the reflector(s), and/or the beam splitter(s), and/or the objective lens(es) and, thus, the multi-dimensional wave field, are rotationally mounted about one or two axes running orthogonally with respect to one another.

23. (New) The wave field microscope as recited in claim 12 wherein provision is made in the detection system for a scanner reflector, which is arranged so as to be suitable for forming an image of the lateral object regions with the desired, preferably maximal, fluorescence intensity.

24. (New) The wave field microscope as recited in claim 12 wherein the illumination system includes in at least one of the three spatial directions, a real illumination source for the two- or multi-photon excitation, and in one or both other spatial direction(s), a real and/or virtual illumination source for the two- or multi-photon excitation, and that the standing wave fields generated with it have wavelengths which differ from one another, and have distances between their specific wave maxima or wave minima of $d_1 = \lambda_1 / 2n \cos \theta_1$ or $d_2 = \lambda_2 / 2n \cos$

θ_2 or $d_i = \lambda_i / 2n \cos \theta_i$ (where: n = the index of refraction in the object space, $\theta_1, \theta_2, \dots, \theta_i$ = the intersection angle of the light wave train of the wavelength $\lambda_1, \lambda_2, \dots, \lambda_i$ with the optical axis), and with the wave fields $WF_1, WF_2 \dots W_i$ being aligned in such away with respect to one another that at least a maximum of two or of all standing waves is situated at the same place (namely the location of a multi-photon excitation).

25. (New) The wave field microscope as recited in claim 12 wherein an arrangement made up of the illumination source, the objective lens, and an electrically conductive reflector, which is suited for generating a one-dimensional, electrical wave field, is provided relative to an object-carrier mount fixture, and, in fact, so as to enable the measuring structures located in the object and/or calibration targets to be aligned through application of the electrical field - prior to or during the microscopic measuring operation.

26. (New) A wave field microscopy method for DNA sequencing, with the use of a wave field microscope comprising:

producing all complementary subsequences of the DNA sequence to be analyzed in such a way that all subsequences begin at the same nucleotide of the sequence to be analyzed;

tagging the fragments to be analyzed at the 3' end with a reference fluorochrome label a and at the 5' end and/or at defined intermediate locations with a fluorochrome label a, g, c, or t — depending on whether the nucleotide base includes adenine (label a), guanine (label g), cytosine (label c) or thymine (label t) -, the fluorochrome labels a, g, c, t and a having different spectral signatures, and each containing one or a plurality of fluorochrome molecules;

fixing the tagged DNA subsequences to a carrier in such a way they are present as a linear sequence, and are placed in a one- or multi-dimensional wave field microscope, with the linear DNA subsequences being so oriented with respect to the standing wave fronts, that a precise distance measurement (accuracy $\pm 1 \cdot 10^{-10}$ m) can be implemented between a and a or g, c or t — once the intensity bary centers are defined and the imaging properties are calibrated -;

registering the signals of the fluorochrome labels step-by-step, spectrally separated from one another;

determining from the distances of spacings between the fluorescent labels and their spectral signatures, the DNA base sequence of the DNA fragment to be analyzed .

27. (New) A calibration method for the multi-dimensional wave field microscopy comprising:

labelling before, during, or after preparing the object in question on or in an object holder, in particular a slide, object carrier fiber, object carrier capillary tube, or object carrier fluid, the object structures to be examined or to be localized - equivalent to the measuring structures - with fluorescent stains having different and/or the same spectral signatures, with such measuring structures to be localized, whose distance from one another is less than the width at half maximum intensity of the effective point spread function, being labeled with fluorescent stains having different spectral signatures;

labelling calibration targets of a defined size and spatial arrangement being with the same fluorescent stains;

preparing the fluorescing calibration targets either together with the objects, i.e. measuring structures, or separately on or in the/ an object holder;

examining the measuring structures and calibration targets microscopically under identical conditions, simultaneously or sequentially;

and in the case of which, two defined calibration targets having different spectral signatures being measured at a time under consideration of the wavelength-dependent imaging and localization properties of the particular optical system, with the measured values ascertained in the process - equivalent to the actual values - being compared to the previously known, actual distance values - equivalent to the reference values -, and from the difference between the actual values and reference values, a correction value - equivalent to the calibration value - being determined, which is used to correct the shift that is conditional upon the optical system, in the detection of various emission loci, in particular of the measuring structures,

the biological object having the fluorochrome-labeled measuring structures, and/or the fluorochrome-labeled calibration target(s), is sequentially or simultaneously illuminated by individual (separate) standing wave fields, running orthogonally to one another in two or three spatial directions, and interfering with one

another to form a two- or three-dimensional wave field, the fluorochromes being excited to emit fluorescence;

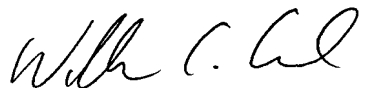
that to detect the fluorescence intensity, a camera and/or one or more two-dimensional arrangement(s) of individual detectors, each having a circular, annular, or slit-shaped plate, or an arrangement of a plurality of circular, annular, or slit-shaped plates is used; and

that either the object having the measuring structures and/or the calibration target(s) or the one- or two-dimensional wave field, or both, is rotated during the measuring operation step-by-step, about one axis or about two axes running orthogonally to one another, the fluorochrome-labeled measuring structures and/or calibration targets being sequentially or simultaneously illuminated by one or two individual standing wave fields disposed orthogonally to one another.

REMARKS

This preliminary amendment is being submitted to conform the specification of the application, which is the national phase of PCT/DE98/01908, to U.S. format. An international search report is submitted herewith. The application is believed to be in condition for allowance and an early review of the application on its merits is hereby respectfully requested. Should the Examiner feel that an interview would advance prosecution of the present application, the Examiner is invited to contact the undersigned.

Respectfully submitted,



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